

# **Cytotoxicity and immunogenicity of SACCHACHITIN and its mechanism of action on skin wound healing**

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摘要

## **Abstract**

SACCHACHITIN membrane, a weavable skin substitute made from the residual fruiting body of *Ganoderma tsugae*, has been demonstrated to promote skin wound healing. Prior to its clinical application, it is critical to learn more about any possible cytotoxicity, immunogenicity, or allergy response, and at least some of its mechanism(s) of action(s). In the present studies, it has been found that SACCHACHITIN suspension at less than 0.05% shows no cytotoxicity to the primary culture of rat fibroblasts. However, at higher concentrations ( $>$  or  $=$  0.1%), it does reduce the growth of fibroblasts, based on MTT assays. This might be caused by positive charges on chitin molecules that are too strong, and may be harmful to the cell membrane.

SACCHACHITIN showed no immunogenicity after it was inoculated into rats three times; however, the unmodified, purified rabbit type I and type II collagens did. Subcutaneous injection of SACCHACHITIN suspension into rats showed no gross allergic responses on skin. Nevertheless, it did cause local acute inflammation, as observed by histological investigation. This is similar to what occurred in the wound site covered with SACCHACHITIN membrane. The chemotactic effect of SACCHACHITIN was exhibited in both intact and wounded skin tissues. This may be one of the initial beneficial effects of SACCHACHITIN membrane to wound healing. The rapid acute inflammatory process was followed by the appearance of angiogenesis and granulation tissue formation, which occurred earlier than it normally would. Coverage of the wound area with SACCHACHITIN membrane also induced an earlier formation of scar tissue to replace the granulation tissue. A 1.5 x 1.5 cm(2) wound area covered by SACCHACHITIN completely healed by 21 days, while that covered with cotton gauze did not. Therefore, SACCHACHITIN is a safe

biomaterial for use as a wound dressing for skin healing. Its promoting action for wound healing might be due to its chemotactic effect for inflammatory cells. This, in turn, may facilitate subsequent angiogenesis, granulation tissue formation, and faster new tissue formation, leading to faster wound healing.